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1: Ann N Y Acad Sci. 1994 May 2;721:208-17. Related Articles, L

Engineering the assembly pathway of the baculovirus-insect cell expressio system.

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The synthesis of complex biological structures such as antibodies using recombinant DN technology is a major biotechnological advance. Active murine antibody (IgG) oligomer composed of two heavy (H) and two light (L) polypeptide chains, have been expressed a secreted by the baculovirus-insect cell expression system. Unfortunately, expression of t functional antibodies is accompanied by the formation of abnormal protein complexes an aggregates in which the polypeptide chains are bound together into incorrect association The formation of these abnormal complexes or protein aggregates in insect cells may be caused by insufficient intracellular levels of two catalytic proteins, immunoglobulin binding protein (BiP or GRP78), and protein disulfide isomerase (PDI). Consequently, w obtained the genes coding for murine BiP and PDI and cloned the genes into the baculovirus vector (Autographa californica nuclear polyhedrosis virus) to obtain AcBB-B and AcBB-PDI. Infection of Spodoptera frugiperda (Sf-9) insect cells with these two baculoviruses yielded recombinant proteins of the correct size that were recognized by antibodies to these proteins. Cloning these genes into the baculovirus vector is one approach to engineering the assembly pathway in order to lower aggregation and increas production of functionally active proteins and oligomers.

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1: Biotechnol Prog. 1991 Sep-Oct;7(5):462-7.

Production of recombinant proteins by baculovirus-infected gypsy moth cells.

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An experimental study was undertaken to evaluate alternative insect cell lines to Sf9 [fro Spodoptera frugiperda (fall armyworm)] for the production of recombinant proteins. Inse cell lines from two different organisms were considered: IPLB-LdEIta (LdEIta) from Lymantria dispar (gypsy moth) and IPLB-HvT1 (HvT1) from Heliothis virescens (tobac budworm). Both LdEIta and HvT1 produced higher total activity levels of recombinant beta-galactosidase in monolayer culture than Sf9 after infection with the Autographa californica nuclear polyhedrosis virus (AcMNPV). However, only LdEIta generated a product yield (activity per milligram of total protein) which exceeded that of Sf9 (by 25% so its growth and production characteristics were investigated in depth. LdEIta generated production levels and yields of a recombinant rotaviral protein, VP4, which exceeded tho of Sf9 by 84 and 38%, respectively. In suspension culture, the LdEIta cells grew as aggregates with a doubling time several hours longer than Sf9, but the recombinant prod yields of LdEIta were still higher than Sf9 by 38% in this culture environment. beta-Galactosidase expression rates and cell death rates suggested that the difference in productivity between the two hosts was due to the ability of LdEIta to survive the baculovirus infection and produce recombinant proteins longer than Sf9. The presence o LdEIta aggregates in suspension culture may be used as a method to separate live cells from dead cells, labile product, and spent medium in recombinant protein production processes.

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